Dear Dr. Li,

Molecules

We would like to re-submit a revised version of our manuscript titled Recent progress of specific G-quadruplex-preferred ligands toward cancer therapy (Manuscript ID: molecules-410205) to this journal.

Based on the reviewers’ comments, we have revised the original text while at the same time maintaining the high standard of the scientific content. We have commented on the modified parts in the revised manuscript. We hope that our revised version is now suitable for publication. The point-by-point responses are described below.

Reviewer 1:

1. l.10 The abstract is too short. Add 2-3 sentences more.
   l.244 expand the abbreviations when appear for the first time.
   l.400 move the figures to the correct positions.

➢ All minor critiques have been reflected in the revised manuscript.

2. I suggest to create the table where the following information will be placed: selective G4-targeting ligands (all or the most important), gene target, cellular/organ/organism effects.

➢ We thank the reviewer for his/her suggestion. We have created the summary table (Table 1) of specific G4-preferred ligands that exhibit antitumor activities.

3. l. 530-537 move the paragraph to the introduction.

➢ We have added the content of this paragraph to the introduction with a small modification;

Relatively recent studies revealed that G4 also had an impact on differentiation- and neuron-related genes [20]. For instance, OCT4 expression may be governed to some degree by G4 formation at the proximal promoter in human embryonic stem cells (CCTL14) [20a], whereas the excessive formation of repetitive G-quadruplex structures
on an expandable (GGGGCC)$_n$ in $C9orf72$ gene or (CGG)$_n$ in $FMRI$ gene accounts for some neurogenetic disorders [20b]. On the contrary, G4 can act positively in neurons, where G4 structures at the CpG island located in x13b are recognized by ATRX, contributing to appropriate synaptic function [20c].

Reviewer 2:

1. The content is interesting and timely but readers with chemistry background involved in drug design and development would expect more information concerning topology of targeted G4 structures, critical comments on binding sites and conclusions related to parameters that decide on the specific binding of ligand to a particular G4 structure. Such data are missing or are scarce.

> We thank the reviewer for his/her comments. Actually, the structure of an EPI-G4 DNA complex was only elucidated at the atomic level among the specific G4-preferred ligands introduced in the manuscript. We have added to the revised manuscript the schematic illustration of the solution structure of the 1:1 EPI-G4 telomere DNA complex whose structure is elucidated by an NMR analysis (Figure 2c).
2. An additional paragraph that summarizes miscellaneous methods and techniques exploited for G4 ligands studies would be appreciated.

- We thank the reviewer for his/her comments. We have added a comment about methods and techniques exploited for G4 ligands studies to Section 2 and cited a suitable reference.

   These pioneering works accelerated the development of G4-selective synthetic molecules, along with the advance in rigid methods and techniques to characterize the binding profiles of such G4 ligands [21a].

3. Other points:
   1. Ref. [82] – I would suggest to replace this ref with other paper of these authors (J. Am. Chem. Soc., 2014, 136 (11), pp 4161–4171)
   2. Figures need more details in captions:
      - Fig. 4: Ref. XX?
      - Fig. 7: caption is before the figure, “molecule” not “molecules”, correct “tageting” to “targeting”, the complex hTERT/ligand GTC365 shown in figure needs more explanation, e.g., meaning of arrows, asterisks and numbers
      - Fig. 9: caption is before the figure
      - Figures 10 and 11 overlapped.

- All minor critiques have been reflected in the revised manuscript.